Thomas M. Jessell et al.

Serial No: 10/002,278

Filed

November 2, 2001

Page 5

posited on October 5, 1992 with the American Type Culture collection (ATCC), 12301 Parklawn Drive, Rockville, Maryland 20852 10801 University Boulevard, Manassas, VA 20110-2209, U.S.A. under the provisions of the Budapest Treaty for the International Recognition of the Deposit of Microorganism for the Purposes of Patent Procedure. Plasmid, pKB 501 (with mycepitope) was accorded ATCC designation number 75320.—

On page 29, lines 21-35, please delete the paragraph which begins "The coding region . . ." and insert the following amended paragraph:

-- The coding region of dorsalin-1 was isolated using the two PCR primers ORF-5' (5' TGGAATTCATCGATAACGGAAGCTGAAGC 3'; SEQ ID 12) and ORF-3' (5' AGCGTCGACATCGATATTCAGCATATACTACC 3'; SEQ ID No. 13) and cloned into pBS SK-between the EcoRI and SalI sites. To insert the c-myc epitope (EQKLISEEDL; SEQ. ID No. 18) two internal primers, each encoding half of the c-myc epitope and dorsalin sequences from the epitope insertion site (see Figure 1 Figures 1A and 1B), were used to produce two PCR fragments, one encoding dorsalin N-terminal to the insertion primer the ORF-5' and (with primer GCGAATTCGATATCAGCTTCTGCTCTGCTCCTATGCTTCTTGC 3' [SEQ. ID No. 14]) and the other encoding the C-terminal region (with primer 5' CGGAATTCGATATCCGAGGAGGACCTGAACCACTGTCGGAGAACGTC 3'; SEQ --

On page 36, lines 1-3, please delete the paragraph which begins "library and to define . . ." and insert the following amended paragraph:

Thomas M. Jessell et al.

Serial No :

10/002,278

Filed

November 2, 2001

Page 6

--library and to define a clone containing a 3.5 kb insert with an open reading frame that encoded a protein of 427 amino acids (Fig. 1 Figures 1A and 1B).-

On page 37, lines 15-34, please delete the paragraph which begins "Medium from cells . . ." and insert the following amended paragraph:

--Medium from cells transfected with the epitope-modified  $dsl ext{-}1$ construct was passed over a MAb 9E10 (Evan et al., 1985) anti Affinity purified proteins were c-myc affinity column. analyzed by gel electrophoresis, revealing a major 15 kDa band and minor bands at 45,47 and -60 kDa (Fig. 3A). The bands at 45 and 47 kDa correspond in size to those predicted for the unproceesed dsl-1 protein and the 15 kDa band to that expected for a proteolytically-cleaved product. To establish the identity of the 15 kDa band and to determine the site for proteolytic cleavage of the precursor protein, the 15 kDa band was blotted onto Immobilon membranes and subjected to sequence The  $\mathrm{NH}_2\text{-terminal}$  sequence obtained, SIGAEQKLIS (SEQ ID No. 16), corresponds to residues 319-322 of the predicted dsl-1 sequence followed by the first 6 residues of the human c-myc epitope. This result shows that the R-S-K-R (SEQ ID No. 17) sequence at residues 315-318 is the site of proteolytic processing of the dsl-1 precursor (arrow in Fig. 1 Figures 1A and 1B), at least in the presence of the c-myc peptide. --

Thomas M. Jessell et al.

Serial No :

10/002,278

Filed

November 2, 2001

Page 7

## In the claims:

Please amend claims 41-48 under the provisions of revised 37 C.F.R.  $\ni 1.121$  as follows:

- --1. 16. (cancelled) --
- --17. (withdrawn) --
- --18. (cancelled) --
- --19. (previously presented) An isolated dorsalin-1 polypeptide comprising continuous amino acids, the sequence of which is set forth in SEQ ID NO: 9.--
- --20. (withdrawn) --
- --21. (cancelled) -
- --22. 27. (withdrawn) --
- --28. 35. (cancelled) --
- --36. 40. (withdrawn) --
- --41. (currently amended) A pharmaceutical composition comprising the isolated dorsalin-1 polypeptide of claim [[17 or]] 19 and a pharmaceutically acceptable carrier.-

Thomas M. Jessell et al.

Serial No :

10/002,278

Filed

November 2, 2001

Page 8

--42. (currently amended) The pharmaceutical composition of claim 41, wherein the pharmaceutical composition comprises an amount of isolated dorsalin-1 polypeptide effective to stimulate neural crest cell differentiation.--

- --43. (currently amended) The pharmaceutical composition of claim 41, wherein the pharmaceutical composition comprises an amount of isolated dorsalin-1 polypeptide effective to regenerate a nerve cell in a subject.--
- --44. (currently amended) The pharmaceutical composition of claim 41, wherein the pharmaceutical composition comprises an amount of isolated dorsalin-1 polypeptide effective to promote bone growth in a subject.--
- --45. (currently amended) The pharmaceutical composition of claim 41, wherein the pharmaceutical composition comprises an amount of isolated dorsalin-1 polypeptide effective to promote wound healing in a subject.-
- --46. (currently amended) The pharmaceutical composition of claim 41, wherein the pharmaceutical composition comprises an amount of isolated dorsalin-1 polypeptide effective to inhibit neural tumor cell growth in a subject.--
- --47. (currently amended) The <del>pharmaceutical</del> composition of claim 46, wherein the neural tumor is a neurofibroma.--

Thomas M. Jessell et al.

Serial No :

10/002,278

Filed

November 2, 2001

Page 9

--48. (currently amended) The pharmaceutical composition of claim 46, wherein the neural tumor is a Schwann cell tumor.--